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Differences in phytoaccumulation of organic pollutants in freshwater submerged and emergent plants

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ABSTRACT

Plants play an important role as sinks for or indicators of semivolatile organic pollutants, however most studies have focused on terrestrial plants and insufficient information has been obtained on aquatic plants to clarify the accumulation of organic pollutants via air-to-leaf vs. water-to-leaf pathways. The presence of *p*, *p'*-dichlorodiphenyldichloroethylene (*p*, *p'*-DDE), hexachlorobenzene (HCB), 15 polycyclic aromatic hydrocarbons (PAHs), and 9 substituted PAHs (*s*-PAHs), including oxy-PAHs and sulfur-PAHs, in 10 submerged and emergent plants collected from Lake Dianchi was analyzed in this study. Relatively low concentrations of *p*, *p'*-DDE (ND to 2.22 ng/g wet weight [ww]) and HCB (0.24–0.84 ng/g ww) and high levels of PAHs (46–244 ng/g ww) and *s*-PAHs (6.0–46.8 ng/g ww) were observed in the aquatic plants. Significantly higher concentrations of most of the compounds were detected in the leaves of the submerged plants than in those of the emergent plants. The percentages of concentration difference relative to the concentrations in the submerged plants were estimated at 55%, 40%, 10%–69% and 0.5%–79% for *p*, *p'*-DDE, HCB, PAHs, and *s*-PAHs, respectively. The percentages were found to increase significantly with an increase in log Kow, suggesting that the high level of phytoaccumulation of pollutants in aquatic plants is due to hydrophobicity-dependent diffusion via the water-to-leaf pathway and the mesophyll morphology of submerged plants.

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1. Introduction

Plants have been shown to be a significant sink for semivolatile organic pollutants due to their surface area and associated lipid content (Paterson et al., 1994; Simonich and Hites, 1994, 1995). The occurrence of pollutants in plant leaves is an important indicator of local contamination (Hermanson and Hites, 1990; Meredith and Hites, 1987; Safe et al., 1992). The phytoaccumulation of organic pollutants in leaves has been widely investigated in previous studies, but attention was paid predominantly to the leaves of terrestrial plants, which accumulate organic pollutants through air-to-leaf pathway (Nakajima et al., 1995; Paterson et al., 1991; Pausch et al., 1995; Simonich and Hites, 1995; Tolls and McLachlan, 1994).

In contrast, aquatic plants were estimated to account for more than 27% of global plants species (Lacoul and Freedman, 2006), and can accumulate organic pollutants through air-to-leaf pathway and water-to-leaf pathways. Exploring the behaviors of pollutants in aquatic plants can help to determine whether aquatic plants are potential biosinks and biofilters of pollutants in aquatic environments, and clarify the accumulation of organic pollutants through air-to-leaf or water-to-leaf pathways in aquatic plant.

Aquatic plants are divided into helophytes, neustophytes and hydrophytes depending on the degree of their immersion in water (Raspopov, 1978; Ronzhina and P'yankov, 2001). Unlike those of terrestrial plants, the leaves of aquatic plants accumulate organic pollutants via both water-to-leaf and air-to-leaf pathways. However, few studies have been conducted on the occurrence of organic pollutants in aquatic plants. These studies mainly investigated emergent plants by assessing the partitioning of pollutants in the roots, stems, and leaves to explore uptake via sediment-to-plant and air-to-plant pathways (Abbassy et al., 2003; Guo et al., 2011;

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Zhou et al., 2008). The importance of air-to-plant pathways could also be identified by comparing the accumulation of organic pollutants in submerged and emergent plants. However, no studies have yet been conducted on the air-to-leaf and water-to-leaf accumulation of organic pollutants in plants, and the differences in the phytoaccumulation of organic pollutants between submerged and emergent plants are still unknown. Therefore, it is vital to analyze pollutant concentrations in the leaves of submerged and emergent plants to compare their phytoaccumulation in these two groups of aquatic plants.

Polycyclic aromatic hydrocarbons (PAHs) have been shown to accumulate in plants (Parrish et al., 2006; Simonich and Hites, 1994). In addition to parent PAHs, substituted PAHs (s-PAHs) such as oxy-PAHs and sulfur-PAHs occur widely in the environment and have various emission sources, such as the reaction of PAHs with atmospheric components and the transformation of PAHs in sediment and soil (Arey et al., 1986; Dimashki et al., 2000; Pitts, 1987). Among the limited studies concerning the environmental occurrences and fates of s-PAHs, no study has been made about their occurrences in plants, although these compounds have been reported to exhibit greater mutagenic and carcinogenic potential than PAHs (Lundstedt et al., 2007).

The present study aimed to assess the bioaccumulation potentials of organic pollutants in aquatic plants, and explore the uptake mechanisms of organic pollutants through air-to-leaf or water-to-leaf pathways. The occurrences of *p*, *p'*-dichlorodiphenyldichloroethylene (*p*, *p'*-DDE), hexachlorobenzene (HCB), PAHs, oxy-PAHs, and sulfur-PAHs in aquatic plants from Lake Dianchi were analyzed. The phytoaccumulation of the targeted pollutants was assessed in both submerged and emergent plants, and the potential mechanisms influencing phytoaccumulation in the two aquatic plant groups were clarified. The findings offer a better understanding of the phytoaccumulation of organic pollutants driven by different ecological processes (air-to-leaf vs. water-to-leaf).

2. Materials and methods

2.1. Chemicals and reagents

The studied chemicals includes *p*, *p'*-DDE, HCB, 15 PAHs (acenaphthylene (Acy), acenaphthene (Ace), fluorene (FE), phenanthrene (Ph), anthracene (An), fluoranthene (Fl), pyrene (Py), chrysene (Ch), benz[a]anthracene (BaA), benzo[b]fluoranthene (BbF), benzo[k]fluoranthene (BkF), benzo[a]pyrene (BaP), indeno [1,2,3-cd]pyrene (IP), benzo[ghi]perylene (BP) and dibenz[a,h]anthracene (DA)), and 9 s-PAHs (dibenzothiophene (DBT), 9-fluorenone (9-Fl), diphenyl disulfide (DD), 2,2'-bithiophene (BT), 1,4-naphthoquinone (NaP), dibenzyl sulfide (DS), benzophenone (BPH), 2-naphthalenethiol (2-NT), thianthrene (TT)). The standards and surrogate standards (PCB 121, acenaphthene-*d*₁₀, phenanthrene-*d*₁₀, chrysene-*d*₁₂, and perylene-*d*₁₄) were purchased from AccuStandard (New Haven, CT). HPLC-graded solvents (dichloromethane, acetonitrile, acetone and hexane) were obtained from Fisher Scientific (NJ). Analytical-graded sodium sulfate and aluminum oxide were purchased from CNW Technologies GmbH (Düsseldorf, Germany) and Sinopharm Chemical Reagent Co., Ltd (China), and heated at 400 °C for 4 h before use.

2.2. Sample collection

Lake Dianchi, a typical shallow plateau lake, is located in Yunnan Province in the middle of the Yungui Plateau of southwestern China. The lake has an elevation of 1886 m, an area of about 300 km² and an average depth of approximately 4.7 m (Hou et al., 2004). The Lake basin is in a northern sub-tropical moist

monsoon climate with average annual precipitation of approximately 1004 mm, average temperature of approximately 14.5 °C, 2470 sunshine hours per year, relative humidity of 74%, and mean wind speed of about 2.5 m/s (Sheng et al., 2012). The lake is heavily contaminated by receiving anthropogenic discharge with total nitrogen and phosphorus of 2.084 and 0.109 mg/L in the water column and 1078 and 327 mg/kg in the sediment. The samples were collected in April 2015 from four littoral zones (24°47'13"N, 102°43'8"E; 24°42'0"N, 102°40'5"E; 25°18'14"N, 102°47'2"E, and 24°51'22"N, 102°46'5"E) in Lake Dianchi. At each sampling location, most the available submerged and emergent plants were collected, and 6–8 individual samples were collected for each species (Table S1). For each species in one location, the plant samples were freeze-dried and homogenized as one sample for chemical analysis. The collected samples included four hydratophytes (whorled watermilfoil [*Myriophyllum verticillatum* L.], potamogetonaceae [*Potamogeton maackianus* A. Bennett], curled pondweed [*Potamogeton crispus* Linn.], and hornwort [*Ceratophyllum demersum* Linn.]), five helophytes (reed [*Phragmites australis* (Cav.) Trin ex Steud.], bulrush [*Typha orientalis* C.Presl], manchurian wild rice [*Zizania latifolia* (Griseb) Stapf], alligator weed [*Alternanthera philoxeroides* (Mart.) Griseb.], and indian lotus [*Nelumbo nucifera* Gaertn.], and one neustophyte (water hyacinth [*Eichhornia crassipes* (Mart.) Solms]).

The aquatic plants were collected, enclosed in aluminum foil, and delivered in an ice-chilled box to a local laboratory. The leaves were selected, cleaned several times with tap water to remove adhering algae and insect larvae, and rinsed with distilled water. Similar steps have been widely described in previous studies investigating pollutants uptake in the plants (Gao et al., 2000; Kim and Kim, 2000; Özer et al., 2006; Waranusantigul et al., 2003). The leaves were cut into small species, homogenized, freeze-dried, and stored at –20 °C in amber glass vials before analysis.

2.3. Sample preparation

The chemical analysis was conducted following the method reported previously by Fan et al. (2017), with some modifications. Briefly, dry samples of approximately 4 g each, spiked with surrogate standards (PCB 121, acenaphthene-*d*₁₀, phenanthrene-*d*₁₀, chrysene-*d*₁₂ and perylene-*d*₁₄), were Soxhlet extracted with 250 mL of dichloromethane and hexane (1:1) solution for 24 h. The resulting extract was treated by acetonitrile partitioning. The extract was rotary-evaporated to nearly 1 mL and then separated in a funnel containing 50 mL of hexane-saturated acetonitrile and 20 mL of hexane. After shaking for 5 min, the bottom acetonitrile layer was collected. Another 50 mL of hexane-saturated acetonitrile was added to the separator funnel and shaken for 5 min. A 100-mL sample of hexane-saturated acetonitrile containing the extract was collected. The acetonitrile extract was concentrated to about 1 mL and further to near-dryness under a gentle gas stream of nitrogen and then re-dissolved in 1 mL hexane. The extract was then passed through a glass column containing 8 g of 5% H₂O-deactivated Al₂O₃. A solution containing 30 mL of hexane and 30 mL of hexane-dichloromethane (3:1) was used to elute the glass column. The eluent was evaporated to about 1 mL and further to near-dryness under a gentle gas stream of nitrogen and then re-dissolved in 0.1 mL hexane for analysis by gas chromatography coupled with mass spectrometry (GC-MS). The sample preparation processes have been validated by recovery experiments.

2.4. GC-MS analysis

The final sample extract was analyzed by GC-MS using a spectrometer (GCMS-QP2010 Plus, Shimadzu) equipped with a fused

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